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Neutrophil and Platelet Activation at Balloon-Injured Coronary Artery Plaque in Patients Undergoing Angioplasty

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Objectives. **This study sought to investigate changes in the expression of activation-dependent adhesion receptors on neutrophils and platelets after exposure to the balloon-injured coronary artery plaque.**

Background. **Activation of blood cells at the balloon-injured coronary artery plaque may contribute to abrupt vessel closure and late restenosis after percutaneous transluminal coronary angioplasty.**

Methods. **In 30 patients undergoing elective coronary angioplasty, blood specimens were obtained through the balloon catheter proximal to the plaque before dilation and distal to the plaque after dilation. Simultaneous blood samples obtained through the guiding catheter served as control samples. Total surface expression of the inducible fibrinogen receptor (CD41) and surface expression of the activated fibrinogen receptor (LIBS1) on platelets as well as Mac-1 (CD11b) and L-selectin (CD62L) surface expression on neutrophils were assessed by flow cytometry.**

Results. **After exposure to the dilated coronary artery plaque, surface expression of LIBS1 on platelets increased by 40.5 ± 11.0 mean (±SE) fluorescence ($p = 0.001$) and that of CD11b on neutrophils increased by 20.1 ± 4.4 mean fluorescence ($p = 0.018$). Concomitantly, anti-CD62L binding on neutrophils decreased by 6.6 ± 2.4 mean fluorescence ($p = 0.022$). In contrast, surface expression of the adhesion receptors did not change significantly between the coronary ostium and the prestenotic coronary segment.**

Conclusions. **The results of this study demonstrate neutrophil and platelet activation at the balloon-injured coronary artery plaque. This cellular activation may serve as a target for pharmacologic interventions to improve the outcome of coronary angioplasty.**

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Balloon angioplasty mechanically disrupts the atherosclerotic plaque and exposes procoagulant and proinflammatory subendothelial structures of the lesion (1,2). Contact activation of the kinin-generating system (3) and presentation of tissue factor contained in the plaque (2) may then induce thrombin formation and platelet activation (4,5). Moreover, leukocytes can be activated by kinins (6) and thrombin (7,8) and thereby attain procoagulant properties (9,10). The resulting thrombus formation may induce abrupt vessel closure and contribute to the proliferative response of vascular cells that causes subsequent restenosis (11,12).

However, in the clinical setting direct evidence for platelet and leukocyte activation at the balloon-injured coronary artery plaque is still lacking. Postmortem studies (13) have demonstrated an accumulation of platelets and white blood cells at the site of vascular injury. Furthermore, examination of blood samples taken from the coronary sinus after angioplasty of the left anterior descending coronary artery revealed transcardiac leukocyte and platelet activation (14–17). Nevertheless, it is unclear whether and to what extent balloon-induced plaque injury contributes to the activation of circulating leukocytes and platelets.

On activation, leukocytes and platelets change surface expression and conformation of adhesion receptors (18,19). With the use of monoclonal antibodies directed against specific epitopes, these changes can be evaluated by flow cytometry in small blood samples. In the present study, we investigated leukocyte and platelet activation at the site of plaque disruption during percutaneous transluminal coronary angioplasty. For this purpose we used flow cytometry to analyze changes in adhesion receptors on platelets and leukocytes that play a key role in the interaction between vascular cells.

Methods

Patients. The study included 30 patients undergoing elective coronary angioplasty for proximal coronary artery stenosis, classified as type A or B (20). Percent diameter stenosis had to be ≥70% on computerized quantitative analysis (21). All patients had Canadian Cardiovascular Society functional class

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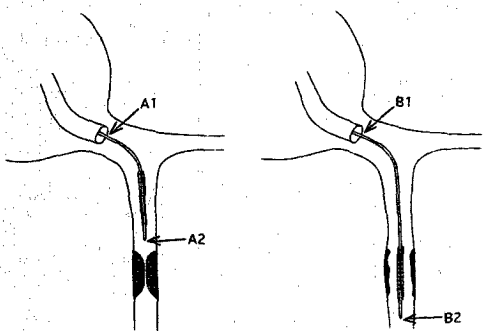


Figure 1. Sampling conditions. Before the first balloon inflation, simultaneous blood samples were obtained through the guiding catheter (A1) and through the balloon catheter placed proximal to the plaque (A2). Immediately after the first balloon inflation, blood samples were drawn simultaneously from the guiding catheter (B1) and from the balloon catheter placed distal to the dilated plaque (B2).

III chronic stable angina. We did not include patients with myocardial infarction within the 3 months preceding the study or those with interfering noncardiac diseases, such as inflammatory disorders, malignancy and infection. The regular medication of the patients was not altered for the study; no patient was taking any anti-inflammatory agent except aspirin 100 mg/day. The study was approved by the Institutional Ethics Committee for Human Subjects. Written informed consent was obtained from all patients.

Study protocol. Angioplasty was performed by the transfemoral approach using 8F guiding catheters with an inner lumen of 0.84 in. (Cordis Corporation) and over the wire balloon catheters with a central lumen of 0.018 in. (Olympix, Cordis Corporation). Before the procedure, 15,000 IU of heparin was given intraarterially and 1 g of aspirin intravenously. With the use of a 0.0014-in. guide wire, the tip of the balloon catheter was placed ~5 mm proximal to the target lesion. After removal of the guide wire, the first set of blood samples was obtained. Blood from the guiding (5 ml) and balloon catheters (0.5 ml) was discarded; 2-ml samples were then drawn simultaneously from the guiding and balloon catheters over a 1-min period (Fig. 1). Thereafter the guide wire was reinserted and the balloon advanced across the lesion (Fig. 1). Balloon inflation was performed for 90 s with sufficient inflation pressure to achieve full expansion (Table 1). During inflation, the guide wire was again removed. Immediately after balloon deflation, we obtained the second set of blood samples by the same protocol as that used for the first set (Fig. 1). Completion of the angioplasty procedure was left to the operator's discretion.

In 10 of the 30 study patients who had moderate coronary artery stenosis (70% to 80%), we took additional blood samples distal to the lesion before dilation and from the coronary ostium. Moreover, in six additional patients we analyzed the influence of the sampling conditions on neutrophil and platelet activation markers. After the preangioplasty medication indicated previously, we obtained blood samples from the abdominal aorta through the guiding catheter alone, the guiding catheter with the balloon catheter inside and the balloon catheter protruding outside the guiding catheter.

Table 1. Baseline and Procedural Characteristics

Age (yr)	
Median	66
Range	36–82
Male/female	19/11
Risk factors	
Hypertension	15
Hypercholesteremia	22
Smoking	16
Diabetes mellitus	4
Medication	
Aspirin	30
Beta-blockers	26
Calcium antagonists	5
ACE inhibitors	14
Diuretic drugs	5
Nitrates	23
Vessel	
LAD	9
LCx	9
RCA	12
Lesion type	
A	10
B	20
Balloon size	
3.0 mm	20
3.5 mm	10
Inflation pressure (atm)	
Median	6
Range	3–9
Dissection after 1st balloon inflation	12

Unless otherwise indicated, data presented are number of patients. ACE = angiotensin-converting enzyme; LAD = left anterior descending coronary artery; LCx = left circumflex coronary artery; RCA = right coronary artery.

Monoclonal antibodies. The following immunoglobulin G murine monoclonal antibodies were used: Bear 1 as anti-CD11b (alpha$_m$ subunit of the beta$_2$-integrin Mac-1); REG56 as anti-CD62L (L-selectin); P2 as anti-CD41, which detects the glycoprotein (GP) IIb-IIIa (inducible platelet fibrinogen receptor) irrespective of whether it is in its rest or activated form (all from Immunotech, Marseille, France); and anti-LIBS1, which recognizes the ligand-induced binding site on GPIIIa (courtesy of Dr. M. Ginsberg, Scripps Clinic, La Jolla, California). All monoclonal antibodies were commercially obtained conjugated with fluoresceine-isothiocyanate except for anti-LIBS1 monoclonal antibody, which was labeled with fluoresceine-isothiocyanate according to standard methods (22).

Flow cytometry and cell counts. For immunofluorescent labeling, the specimens were anticoagulated with 1:5 (vol/vol) CPDA (sodium citrate, phosphate buffer, dextrose, adenine; Fa. Greiner, Nürtingen, Germany) and processed immediately after sampling. Leukocyte staining was performed in whole-

Table 2. Coronary Artery–Ostium Differences in Cell Count and Expression of Adhesion Receptors

	Across Prestenotic CA Segment (n = 30)		Across Dilated CA Segment (n = 30)	
	Median (quartiles)	p Value*	Median (quartiles)	p Value*
Neutrophils				
Count ($\times 10^9$/liter)	0.04 (−0.12, 0.28)	0.45	0.03 (−0.12, 0.3)	0.43
CD11b (mean fl.)	2.0 (−2.2, 5.9)	0.27	18.2 (3.6, 32.4)	0.018
CD62L (mean fl.)	1.1 (−4.1, 2.8)	0.89	−5.1 (−11.0, 2.4)	0.022
Platelets				
Count ($\times 10^9$/liter)	5.5 (−10.5, 21)	0.88	−7.0 (−11, 24.5)	0.91
LIBS1 (mean fl.)	−1 (−27, 19)	0.40	23 (2, 72)	0.001
CD41 (mean fl.)	0.5 (−6, 9)	0.61	1.5 (−9, 10)	0.70

*Level of significance for coronary artery (CA)–ostium values different from zero. fl. = fluorescence.

blood samples (23). We added 25 μl of whole blood and an equal volume of phosphate-buffered saline containing saturating concentrations of anti-CD11b or anti-CD62L monoclonal antibodies. After incubation for 30 min at room temperature, erythrocytes were lysed and leukocytes were fixed using commercially available solutions (Immunolyse and Fixative, Coulter Electronics, Krefeld, Germany). Thereafter, the cells were washed three times and stored in 1% paraformaldehyde at 4°C in the dark.

Platelet immunostaining was performed as described elsewhere (22,24). Briefly, 5 μl of platelet-rich plasma was incubated with saturating concentrations of anti-CD41 and anti-LIBS1 monoclonal antibodies for 30 min at room temperature. Samples were then diluted with 2 ml 0.5% paraformaldehyde and stored at 4°C. Flow cytometric analysis of the fixed samples was performed within 24 h after sampling.

Antibody binding was assessed by flow cytometry using a FACScan (Becton-Dickinson) equipped with a 488-nm argon laser (500 mW) (22–24). Fluorescent monosized beads (Cali-BRITE, Becton-Dickinson) were used to check the reproducibility of the method. Neutrophils and platelets were identified by their characteristic forward angle and right-angle light-scattering properties. Fluorescence intensity (mean fluorescence) of 5,000 neutrophils or platelets was recorded as a mean channel number using a logarithmic scale. Data were stored on list mode files and processed on a Hewlett-Packard computer equipped with scientific software. In healthy volunteers, the day to day variability of the immunofluorescence measurements ranged from 1% to 5% for CD11b and CD41 and from 8% to 14% for CD62L and LIBS1.

Whole-blood cell counts were performed in ethylenediaminetetraacetic acid–anticoagulated (1 g/liter) specimens using a Sysmex Counter (Digitana, Model F800, Hamburg, Germany).

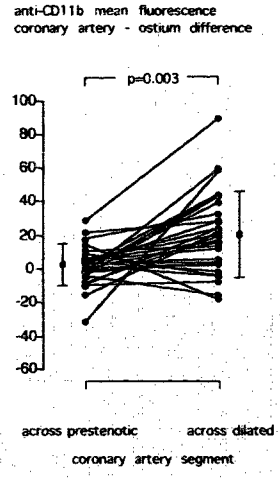
Statistical analysis. The Kolmogorov-Smirnov test showed that the data were not normally distributed. Thus, results are reported as median (quartiles), unless otherwise indicated. The differences between matched samples were tested by the Wilcoxon matched-pairs signed-rank test, and those between two groups by the Mann-Whitney *U* test. A p value <0.05 in the two-tailed test was regarded as significant.

Results

Clinical and angiographic data. Table 1 shows baseline characteristics of the patients and their regular medication as well as angiographic data and procedural characteristics. The first balloon inflation achieved a significant reduction of the stenosis (percent diameter stenosis <50%), and contrast runoff was unimpaired. Dissection was visible in 12 patients. No acute or subacute closures of the dilated vessel occurred.

Surface expression on leukocytes of CD11b and CD62L. Surface expression of CD11b on neutrophils sampled downstream of the dilated plaque was significantly higher than that in ostial samples (Table 2, Fig. 2). Concomitantly, we found a diminished anti-CD62L immunofluorescence on neutrophils obtained downstream of the dilated plaque compared with

Figure 2. Individual coronary artery–ostium differences (**circles**) in CD11b surface expression on neutrophils across prestenotic and dilated coronary artery segments. Mean value (**squares**) ± SD (**vertical bars**) of data points are also shown; p value indicates level of significance for difference between values before and after angioplasty.



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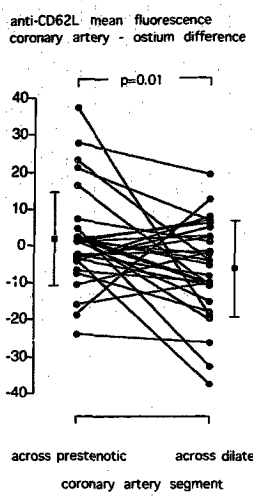

Figure 3. Individual coronary artery–ostium differences **(circles)** in CD62L surface expression on neutrophils across prestenotic and dilated coronary artery segments. Mean value **(squares)** ± SD **(vertical bars)** of data points are also shown; p value indicates level of significance for difference between values before and after angioplasty.

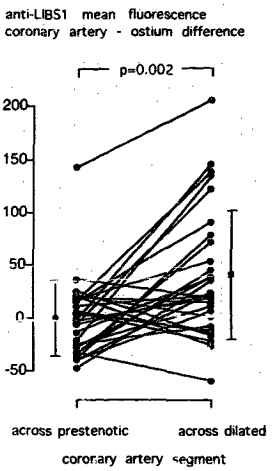

Figure 4. Individual coronary artery–ostium differences **(circles)** in LIBS1 surface expression on platelets across prestenotic and dilated coronary artery segments. Mean value **(squares)** ± SD **(vertical bars)** of data points are also shown; p value indicates level of significance for difference between values before and after angioplasty.

samples from the coronary ostium (Table 2, Fig. 3). On the contrary, in samples obtained proximal to the lesion before dilation, surface expression of CD11b and CD62L on neutrophils did not differ significantly from coronary ostial samples (Table 2, Fig. 2 and 3). When the two sampling conditions (proximal to the lesion before and distal to the lesion after angioplasty) were compared, the difference between the coronary artery and the ostium in anti-CD11b immunofluorescence increased significantly and that in anti-CD62L immunofluorescence decreased significantly (Fig. 2 and 3).

In the 10 patients in whom we obtained blood samples distal to the nondilated lesion, we did not find significant gradients in the surface expression of CD11b and CD62L on neutrophils between the downstream vicinity of the nondilated plaque and the coronary ostium (median difference in anti-CD11b mean fluorescence 1.2 [interquartile range −10.1 to 8.0], p = 0.88; median difference in anti CD62L mean fluorescence 0.1 [interquartile range −1.9 to 5.7], p = 0.72). We did not find significant coronary artery–ostium differences in leukocyte counts either before or after angioplasty (Table 2).

Effect of angioplasty on platelet fibrinogen receptor surface expression. Similar to CD11b expression on neutrophils, anti-LIBS1 immunofluorescence on platelets sampled downstream of the dilated plaque was significantly higher than that in ostial samples (Table 2, Fig. 4). However, platelet anti-LIBS1 immunofluorescence did not differ significantly between coronary artery specimens sampled proximal to the lesion before dilation and ostial specimens (Table 2, Fig. 4). Thus, the coronary artery–ostium difference in anti-LIBS1 immunofluorescence increased significantly between sampling proximal to the lesion before and distal to the lesion after angioplasty (see Fig. 4). The increase in anti-LIBS1 immunofluorescence downstream of the dilated plaque was not associated with an increase in anti-CD41 binding, nor did we find coronary artery–ostium differences in anti-CD41 immunofluorescence before angioplasty (Table 2). Moreover, platelet counts did not show significant changes (Table 2).

In the subgroup with moderate coronary artery stenosis, we did not find significant differences in anti-LIBS1 mean fluorescence (median 0 [interquartile range −15 to 12], p = 1.0) between the coronary ostium and the downstream vicinity of the nondilated coronary artery lesion.

Relation between platelet and neutrophil membrane markers and angiographic variables. Surface expressions of CD11b or CD62L on neutrophils and those of CD41 and LIBS1 on platelets were not significantly related to any of the procedural or angiographic variables shown in Table 1. Nevertheless, if dissection was visible after the first balloon inflation, there was a trend toward a larger difference in CD11b immunofluorescence between neutrophils sampled downstream of the dilated plaque and those from ostial samples (Table 3).

Sampling through different catheters. We did not find significant differences in surface expression of adhesion receptors between samples from the abdominal aorta obtained through the balloon catheter and those obtained through the guiding catheter with the balloon catheter inside (Table 4). Similarly, anti-CD11b and anti-CD62L immunofluorescence on neutrophils or anti-LIBS1 and anti-CD41 immunofluorescence on platelets did not differ between samples obtained through the guiding catheter with the balloon catheter inside

Table 3. Coronary Artery–Ostium Differences Across Dilated Coronary Artery Segment in Expression of Adhesion Receptors in Patients With and Without Visible Dissection

	With Dissection (n = 12)	Without Dissection (n = 18)	p Value
CD11b (mean fl.)	61.6 (17.0, 98.5)	21.5 (3.6, 56.8)	0.11
CD62L (mean fl.)	−4.0 (−10.5, 4.0)	−7.5 (−16.0, 2.0)	0.63
LIBS1 (mean fl.)	29 (9.5, 102)	22 (−11, 45)	0.46

Data presented are median (quartiles). fl. = fluorescence.

and those obtained through the guiding catheter alone (data not shown).

Discussion

The results of this study present evidence of neutrophil and platelet activation at the balloon-injured coronary artery plaque. After exposure to the dilated coronary artery segment, neutrophils and platelets undergo functional changes manifested by a significant increase in the surface expression of LIBS1 on platelets and of CD11b on neutrophils. Thus, after exposure to the dilated coronary artery segment, platelets activated their inducible fibrinogen receptors and neutrophils upregulated Mac-1 receptors on their surface. Concomitantly, as another consequence of activation (18,23), neutrophils shedded L-selectin. None of these changes occurred across the nonstenotic coronary artery segment nor across the stenotic nondilated coronary artery segment. These findings suggest that balloon injury to the plaque is an essential prerequisite for the observed changes in adhesion receptor expression on neutrophils and platelets.

Because we did not find differences in neutrophil or platelet count across the dilated coronary artery segment, our findings cannot be explained by changes in cell population. Therefore, the observed changes in surface markers on neutrophils and platelets indicate cell activation (18,19). Potential underlying mechanisms include contact activation of platelets by exposed collagen fibers and formation of mediators at the balloon-injured plaque, in particular kinins and thrombin (3–5). Moreover, atherosclerotic plaques contain activated T cells, monocytes and macrophages that produce proinflammatory cytokines such as tumor necrosis factor-alpha (25,26). These cytokines as well as other macrophage-derived mediators (25) may be released on plaque rupture during angioplasty.

Table 4. Differences in Adhesion Receptor Expression Between Arterial Samples Obtained Through Balloon Catheter and Guiding Catheter With Balloon Catheter Inside

	Difference in Surface Expression (mean fl. ± SE)	p Value*
Neutrophils		
CD11b	−1.4 ± 4.3	0.75
CD62L	1.6 ± 3.5	0.75
Platelets		
LIBS1	5.0 ± 11.2	0.92
CD41	−1.8 ± 6.8	0.83

*Level of significance for values different from zero. fl. = fluorescence.

Comparison with previous studies. Previous studies have examined changes in neutrophil and platelet function in the coronary sinus blood during balloon angioplasty (14–17). However, this approach is likely to miss subtle changes across the dilated coronary artery segment, owing to the substantial admixture of blood from nondilated vessels. In keeping with these considerations, we were unable to detect transcardiac gradients in surface expression on neutrophils of Mac-1 and L-selectin during elective angioplasty (23).

Nevertheless, other studies have described an increase in the surface expression of the activated fibrinogen receptor and hyperaggregation of platelets from the coronary sinus blood as compared with the arterial blood (14,25) as well as transcardiac changes in neutrophil elastase release and superoxide anion production after angioplasty (16,17). However, transcardiac gradients in cell function do not allow characterization of the site of cellular activation. In a previous study (16) we presented evidence that some of the cardiac changes in neutrophil function during angioplasty are related to ischemia and thus are likely to occur within coronary microcirculation. Our present findings suggest that platelet and neutrophil activation at the balloon-injured plaque is an additional mechanism for the transcardiac changes in blood cell function during angioplasty.

Study limitations. The surface expression of the receptors studied may be affected by shear along the outside of the balloon catheter within the narrow confines of the coronary artery. However, the first sampling across the segment without obstructive coronary artery disease did not show any changes in surface expression of adhesion receptors between the coronary artery and the coronary ostium. After successful angioplasty, shear conditions in the dilated segment may not be substantially different from those in nonstenotic segments. Therefore, we cannot assume that shear conditions are a major determinant of the observed changes in platelet and neutrophil function. This interpretation is supported by the analysis of blood samples obtained distal to the lesion before dilation. In these samples we could not detect differences in adhesion receptor expression between the coronary artery and the coronary ostium. This data set was obtained in 10 patients with moderate grade coronary artery stenosis. In the other patients we were concerned that the balloon catheters with profiles of 0.032 to 0.033 in. substantially compromised coronary blood flow, often causing complete obstruction, when placed within high grade stenoses.

Even though the increases in Mac-1 expression on neutrophils by trend were most pronounced at plaques with visible dissection after angioplasty, we could not relate the changes in neutrophil and platelet function to any of the procedural or angiographic variables. This may reflect the inability of angiography to assess the plaque injury with sufficient accuracy.

It has been shown (14) that in patients treated with heparin

and aspirin, the platelet responses in samples drawn through 7F catheters do not differ from those obtained by venipuncture. Nevertheless, we were concerned about artifacts introduced by the sampling procedure. Therefore, we performed control experiments to check potential differences in membrane markers after sampling through the guiding catheter or through the balloon catheter. These experiments showed that the two sampling conditions do not exert an appreciable differential effect on membrane markers. Thus, the methods described herein are suitable for studying cellular activation in blood samples obtained through long catheters.

Pathophysiologic implications and clinical perspectives. Neutrophils that are activated at the balloon-injured coronary artery plaque may contribute to microvascular impairment after angioplasty by increased interaction with the vessel wall and by release of proteolytic enzymes, oxygen-free radicals and vasoactive arachidonic acid metabolites (16,27,28). When recruited to the atherosclerotic lesion, they may aggravate plaque injury (28). Moreover, it is tempting to speculate that the inflammatory stimuli at the balloon-injured plaque may initiate systemic responses. If considered as a paradigm of plaque injury, the findings of the present study may help to explain the systemic inflammatory response syndrome (29), commonly found in acute coronary syndromes (25,30). As shown recently (30), the extent of these systemic inflammatory responses is related to an adverse clinical outcome.

Although we were unable to detect significant differences in platelet count across the dilated coronary artery segment, it is known that platelets are deposited at the site of balloon injury (4,13). Under adverse circumstances this can lead to abrupt vessel closure. Moreover, experimental and recent clinical studies (11,12) suggest that platelet deposition at the dilated plaque contributes to the development of restenosis. Approximately 70% of platelet deposition at the site of balloon injury is fibrinogen receptor dependent (4). Our findings demonstrate that despite treatment with heparin and aspirin, the inducible fibrinogen receptor on platelets is activated at the balloon-injured coronary artery plaque. Therefore, the present study adds weight to recent pharmacologic developments aimed at improving the clinical outcome of angioplasty by blocking platelet fibrinogen receptors.

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References

1. Steele PM, Chesebro JH, Stanson AW, et al. Balloon angioplasty: natural history of the pathophysiological response to injury in a pig model. Circ Res 1985;57:105–12.
2. Wilcox JN, Smith KM, Schwartz S, Gordon D. Localization of tissue factor in the normal vessel wall and in the atherosclerotic plaque. Proc Natl Acad Sci USA 1986;134:1087–97.
3. Scott CF, Purdon AD, Silver LD, Colman RW. Cleavage of high molecular weight kininogen (HMWK) by plasma factor XIa. J Biol Chem 1985;260: 10856–63.
4. Kaplan AV, Leung LL, Leung WH, Grant GW, McDougall IR, Fischell TA. Roles of thrombin and platelet membrane glycoprotein IIb/IIIa in platelet-subendothelial deposition after angioplasty in an ex vivo whole artery model. Circulation 1991;84:1279–88.
5. Meloni FJ, Schmaier AH. Low molecular weight kininogen binds to platelets to modulate thrombin-induced platelet activation. J Biol Chem 1991;266: 6786–94.
6. Regoli D, Barabe J. Pharmacology of bradykinin and related kinins. J Pharmacol Rev 1980;32:1–45.
7. Bizios R, Lai L, Fenton JW II, Malik AB. Thrombin-induced chemotaxis and aggregation of neutrophils. J Cell Physiol 1986;128:485–90.
8. Bar-Shavit R, Kahn A, Wildner GD, Fenton JW II. Monocyte chemotaxis: stimulation by specific exosite region in thrombin. Science 1983;220:728–31.
9. Ruf W, Edgington TS. Structural biology of tissue factor, the initiator of thrombogenesis in vivo. FASEB J 1994;8:385–90.
10. Edwards RL, Rickles FR. The role of leukocytes in the activation of blood coagulation. Semin Hematol 1992;29:202–12.
11. Ip JH, Fuster V, Israel D, Badimon L, Badimon J, Chesebro JH. The role of platelets, thrombin and hyperplasia in restenosis after coronary angioplasty. J Am Coll Cardiol 1991;17:77B–88B.
12. Liu MW, Roubin GS, King SB. Restenosis after coronary angioplasty. Potential biologic determinants and role of intimal hyperplasia. Circulation 1989;79:1374–87.
13. Block PC, Myler RK, Stertzer S, Fallon JR. Morphology after transluminal angioplasty in human beings. N Engl J Med 1982;305:382–6.
14. Gasperetti CM, Gonias SL, Gimple LW, Powers ER. Platelet activation during coronary angioplasty in humans. Circulation 1993;88:2728–34.
15. Schart RE, Tomer A, Marzec UM, Teirstein PS, Ruggeri ZM, Harker LA. Activation of platelets in blood perfusing angioplasty-damaged coronary arteries. Arterioscler Thromb Vasc Biol 1992;12:1475–87.
16. Neumann FJ, Richardt G, Schneider M, et al. Cardiac release of chemoattractants after ischaemia induced by coronary balloon angioplasty. Br Heart J 1993;70:27–34.
17. De Servi S, Mazzone A, Ricevuti G, et al. Granulocyte activation after coronary angioplasty in humans. Circulation 1990;82:140–6.
18. Kuijpers TW, Tool ATJ, Van der Schoot K, et al. Membrane surface antigen expression on neutrophils: a reappraisal of the use of surface markers for neutrophil activation. Blood 1991;78:1105–11.
19. Shattil SJ, Cunningham M, Hoxie JA. Detection of activated platelets in whole blood using activation-dependent monoclonal antibodies and flow cytometry. Blood 1987;70:307–15.
20. Ellis SC, Vandormael SC, Cowley MJ, et al. Coronary morphologic and clinical determinants of procedural outcome with angioplasty for multivessel disease. Circulation 1990;82:1193–203.
21. Kastrati A, Schömig A, Dietz R, Neumann FJ, Richardt G. Time course of restenosis during the first year after emergency coronary stenting. Circulation 1993;87:1498–505.
22. Gawaz MP, Dobos G, Späth M, Mujais SK. Platelet membrane glycoprotein GPIIb-IIIa function is impaired in endstage renal failure. J Am Soc Nephrol 1994;5:36–46.
23. Neumann FJ, Ott I, Gawaz M, et al. Cardiac release of cytokines and inflammatory responses in acute myocardial infarction. Circulation 1995;92: 748–55.
24. Gawaz MP, Neumann FJ, Ott I, Schiessler A, Schömig A. Platelet function in acute myocardial infarction treated by direct angioplasty. Circulation 1996;93:229–37.
25. Alexander RW. Inflammation and coronary artery disease. N Engl J Med 1994;331:468–9.
26. Barath P, Fishbein MC, Coa J, Berenson J, Helfant RH, Forrester JF. Detection and localization of tumor necrosis factor in human atheroma. Am J Cardiol 1990;65:297–302.
27. Entman ML, Ballantyne CM. Inflammation in acute coronary syndromes. Circulation 1993;88:800–3.
28. Westlin WF, Gimbrone MAJ. Neutrophil-mediated damage to human vascular endothelium. Role of cytokine activation. Am J Pathol 1993;142: 117–28.
29. Bone RC. Toward an epidemiology and natural history of SIRS (systemic inflammatory response syndrome). JAMA 1992;268:3452–5.
30. Liuzzo G, Biasucci L, Gallimore J, et al. The prognostic value of C-reactive protein and serum amyloid A protein in severe unstable angina. N Engl J Med 1994;331:417–24.